

1

**DIPEPTIDYL PEPTIDASE INHIBITORS****RELATED APPLICATION**

This application claims the benefit of U.S. Provisional Application No. 60/553,571 filed Mar. 15, 2004 and U.S. Provisional Application No. 60/629,524 filed Nov. 18, 2004, each of which is incorporated herein by reference.

**FIELD OF THE INVENTION**

The invention relates to compounds that may be used to inhibit dipeptidyl peptidases as well as compositions of matter and kits comprising these compounds. The present invention also relates to methods for inhibiting dipeptidyl peptidases as well as treatment methods using compounds according to the present invention.

**DESCRIPTION OF RELATED ART**

Dipeptidyl Peptidase IV (IUBMB Enzyme Nomenclature EC.3.4.14.5) is a type II membrane protein that has been referred to in the literature by a wide variety of names including DPP4, DP4, DAP-IV, FAP $\beta$ , adenosine deaminase complexing protein 2, adenosine deaminase binding protein (ADAbp), dipeptidyl aminopeptidase IV; Xaa-Pro-dipeptidyl-aminopeptidase; Gly-Pro naphthylamidase; postproline dipeptidyl aminopeptidase IV; lymphocyte antigen CD26; glycoprotein GP110; dipeptidyl peptidase IV; glycylproline aminopeptidase; glycylproline aminopeptidase; X-prolyl dipeptidyl aminopeptidase; pep X; leukocyte antigen CD26; glycylprolyl dipeptidylaminopeptidase; dipeptidyl-peptide hydrolase; glycylprolyl aminopeptidase; dipeptidyl-aminopeptidase IV; DPP IV/CD26; amino acyl-prolyl dipeptidyl aminopeptidase; T cell triggering molecule Tp103; X-PDAP. Dipeptidyl Peptidase IV is referred to herein as "DPP-IV."

DPP-IV is a non-classical serine aminodipeptidase that removes Xaa-Pro dipeptides from the amino terminus (N-terminus) of polypeptides and proteins. DPP-IV dependent slow release of dipeptides of the type X-Gly or X-Ser has also been reported for some naturally occurring peptides.

DPP-IV is constitutively expressed on epithelial and endothelial cells of a variety of different tissues (intestine, liver, lung, kidney and placenta), and is also found in body fluids. DPP-IV is also expressed on circulating T-lymphocytes and has been shown to be synonymous with the cell-surface antigen, CD-26. DPP-IV has been implicated in a number of disease states, some of which are discussed below.

DPP-IV is responsible for the metabolic cleavage of certain endogenous peptides (GLP-1 (7-36), glucagon) in vivo and has demonstrated proteolytic activity against a variety of other peptides (GHRH, NPY, GLP-2, VIP) in vitro.

GLP-1 (7-36) is a 29 amino-acid peptide derived by post-translational processing of proglucagon in the small intestine. GLP-1 (7-36) has multiple actions in vivo including the stimulation of insulin secretion, inhibition of glucagon secretion, the promotion of satiety, and the slowing of gastric emptying. Based on its physiological profile, the actions of GLP-1 (7-36) are believed to be beneficial in the prevention and treatment of type II diabetes and potentially obesity. For example, exogenous administration of GLP-1 (7-36) (continuous infusion) in diabetic patients has been found to be efficacious in this patient population. Unfortunately, GLP-1 (7-36) is degraded rapidly in vivo and has been shown to have a short half-life in vivo ( $t_{1/2}$ =1.5 minutes).

Based on a study of genetically bred DPP-IV knock out mice and on in vivo/in vitro studies with selective DPP-IV

2

inhibitors, DPP-IV has been shown to be the primary degrading enzyme of GLP-1 (7-36) in vivo. GLP-1 (7-36) is degraded by DPP-IV efficiently to GLP-1 (9-36), which has been speculated to act as a physiological antagonist to GLP-1 (7-36). Inhibiting DPP-IV in vivo is therefore believed to be useful for potentiating endogenous levels of GLP-1 (7-36) and attenuating the formation of its antagonist GLP-1 (9-36). Thus, DPP-IV inhibitors are believed to be useful agents for the prevention, delay of progression, and/or treatment of conditions mediated by DPP-IV, in particular diabetes and more particularly, type 2 diabetes mellitus, diabetic dislipidemia, conditions of impaired glucose tolerance (IGT), conditions of impaired fasting plasma glucose (IFG), metabolic acidosis, ketosis, appetite regulation and obesity.

DPP-IV expression is increased in T-cells upon mitogenic or antigenic stimulation (Mattem, T., et al., *Scand. J. Immunol.*, 1991, 33, 737). It has been reported that inhibitors of DPP-IV and antibodies to DPP-IV suppress the proliferation of mitogen-stimulated and antigen-stimulated T-cells in a dose-dependant manner (Schon, E., et al., *Biol. Chem.*, 1991, 372, 305). Various other functions of T-lymphocytes such as cytokine production, IL-2 mediated cell proliferation and B-cell helper activity have been shown to be dependent on DPP-IV activity (Schon, E., et al., *Scand. J. Immunol.*, 1989, 29, 127). DPP-IV inhibitors, based on boroProline, (Flentke, G. R., et al., *Proc. Nat. Acad. Sci. USA*, 1991, 88, 1556) although unstable, were effective at inhibiting antigen-induced lymphocyte proliferation and IL-2 production in murine CD4+ T-helper cells. Such boronic acid inhibitors have been shown to have an effect in vivo in mice causing suppression of antibody production induced by immune challenge (Kubota, T. et al., *Clin. Exp. Immun.*, 1992, 89, 192). The role of DPP-IV in regulating T lymphocyte activation may also be attributed, in part, to its cell-surface association with the transmembrane phosphatase, CD45. DPP-IV inhibitors or non-active site ligands may possibly disrupt the CD45-DPP-IV association. CD45 is known to be an integral component of the T-cell signaling apparatus. It has been reported that DPP-IV is essential for the penetration and infectivity of HIV-1 and HIV-2 viruses in CD4+ T-cells (Wakselman, M., Nguyen, C., Mazaleyrat, J.-P., Callebaut, C., Krust, B., Hovanessian, A. G., Inhibition of HIV-1 infection of CD 26+ but not CD 26-cells by a potent cyclopeptidic inhibitor of the DPP-IV activity of CD 26. Abstract P.44 of the 24.sup.th European Peptide Symposium 1996). Additionally, DPP-IV has been shown to associate with the enzyme adenosine deaminase (ADA) on the surface of T-cells (Kameoka, J., et al., *Science*, 193, 26 466). ADA deficiency causes severe combined immunodeficiency disease (SCID) in humans. This ADA-CD26 interaction may provide clues to the pathophysiology of SCID. It follows that inhibitors of DPP-IV may be useful immunosuppressants (or cytokine release suppressant drugs) for the treatment of among other things: organ transplant rejection; autoimmune diseases such as inflammatory bowel disease, multiple sclerosis and rheumatoid arthritis; and the treatment of AIDS.

It has been shown that lung endothelial cell DPP-IV is an adhesion molecule for lung-metastatic rat breast and prostate carcinoma cells (Johnson, R. C., et al., *J. Cell. Biol.*, 1993, 121, 1423). DPP-IV is known to bind to fibronectin and some metastatic tumor cells are known to carry large amounts of fibronectin on their surface. Potent DPP-IV inhibitors may be useful as drugs to prevent metastases of, for example, breast and prostate tumors to the lungs.

High levels of DPP-IV expression have also been found in human skin fibroblast cells from patients with psoriasis, rheumatoid arthritis (RA) and lichen planus (Raynaud, F., et al., *J.*